

Efficiency of Sampling and Analysis of Asbestos Fibers on Filter Media: Implications for Exposure Assessment

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To measure airborne asbestos and other fibers, an air sample must represent the actual number and size of fibers. Typically, mixed cellulose ester (MCE, 0.45 or 0.8 μm pore size) and, to a much lesser extent, capillary-pore polycarbonate (PC, 0.4 μm pore size) membrane filters are used to collect airborne asbestos for count measurement and fiber size analysis. In this research study, chrysotile asbestos (fibers both shorter and longer than 5 μm) were generated in an aerosol chamber and sampled by 25 mm diameter MCE filter media to compare the fiber retention efficiency of 0.45 μm pore size filters vs. 0.8 μm pore size filter media. In addition, the effect of plasma etching times on fiber densities was evaluated. This study demonstrated a significant difference in fiber retention efficiency between 0.45 μm and 0.8 μm pore size MCE filters for asbestos aerosols (structures longer than or equal to 0.5 μm length). The fiber retention efficiency of a 0.45 μm pore size MCE filter is statistically significantly higher than that of the 0.8 μm pore size MCE filter. However, for asbestos structures longer than 5 μm , there is no statistically significant difference between the fiber retention efficiencies of the 0.45 μm and 0.8 μm pore size MCE filters. The mean density of asbestos fibers (longer than or equal to 0.5 μm) increased with etching time. Doubling the etching time increased the asbestos filter loading in this study by an average of 13%. The amount of plasma etching time had no effect on the filter loading for fibers longer than 5 μm . Many asbestos exposure risk models attribute health effects to fibers longer than 5 μm . In these models, both the 0.45 μm and 0.8 μm pore size MCE filter can produce suitable estimates of the airborne asbestos concentrations. However, some models suggest a more significant role for asbestos fibers shorter than 5 μm . Exposure monitoring for these models should consider only the 0.45 μm pore size MCE filters as recommended by the U.S. Environmental Protection Agency Asbestos Hazard Emergency Response Act (AHERA) protocol and other methods.

Keywords asbestos, exposure, filters, microscopy, sampling

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INTRODUCTION

Numerous optical and electron microscopy methods have been developed to detect and quantify asbestos in air, as well as in other matrices. Each method has its own strengths and weaknesses, and they must be carefully evaluated to determine how best to detect and quantify asbestos under a given circumstance.⁽¹⁻¹⁰⁾

Typically, mixed cellulose ester (0.45 μm or 0.8 μm pore size) and, to a lesser extent, capillary-pore polycarbonate (0.4 μm pore size) membrane filters are used to collect airborne asbestos for count measurement and fiber size analysis. It is important to recognize that pore size specification for a membrane filter is an absolute specification only for capillary-pore type filters such as the polycarbonate (PC). The pore size rating for tortuous path filters such as the mixed-cellulose ester (MCE) filters, is an effective pore size and not a specification that particles exceeding that size are retained by the filter.⁽¹¹⁾

The two types of filters differ in their chemical and physical composition. Polycarbonate filters have a smooth filtering surface; the pores are cylindrical, almost uniform in diameter, and essentially perpendicular to the surface (Figure 1). A mixed cellulose ester filter is a thicker filter with a spongelike appearance, and it relies on a tangled maze of cellulose ester strands to trap fibers (Figure 2). For microscopic analysis of asbestos deposited on the filter, it is critical that the fibers be in a single plane to ensure they are in focus during the analysis. This requirement is simple to achieve for PC filters because of the smooth filtering surface, whereas the MCE filter requires two additional steps in the direct preparation procedure. The MCE filter must be collapsed with an organic solvent, and then the top layer of the collapsed filter material must be etched away with a low-temperature plasma asher.

The U.S. Environmental Protection Agency (EPA)⁽¹⁾ and the National Institute for Occupational Safety and Health (NIOSH)⁽⁵⁾ recommend using 0.45 μm pore size MCE filters

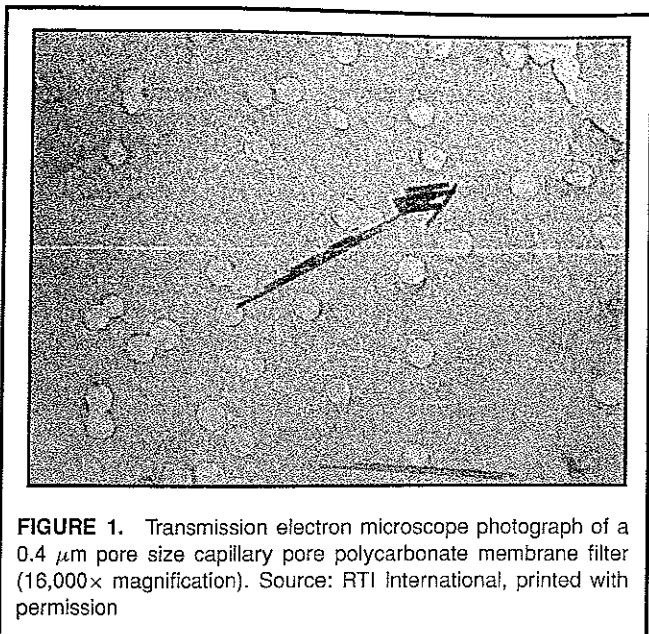


FIGURE 1. Transmission electron microscope photograph of a 0.4 μm pore size capillary pore polycarbonate membrane filter (16,000 \times magnification). Source: RTI International, printed with permission

when performing transmission electron microscopy (TEM) analysis on the samples because the particles deposit closer to the surface than in larger pore size (e.g., 0.8 μm pore size) MCE filters. However, the higher pressure drop through the 0.45 μm pore size MCE filters normally preclude their use with battery-powered personal sampling pumps.⁽⁵⁾

To obtain a uniform distribution of collected particles across the surface of the collecting filter, The U.S. EPA⁽¹⁾ requires a 5.0 μm pore size MCE backing filter be placed behind the collecting filter followed by a cellulose support. This tandem filter assembly further increases the pressure drop, which at given velocity is directly proportional to the thickness of the filter. ISO Method 10312:1995 also recommends the tandem

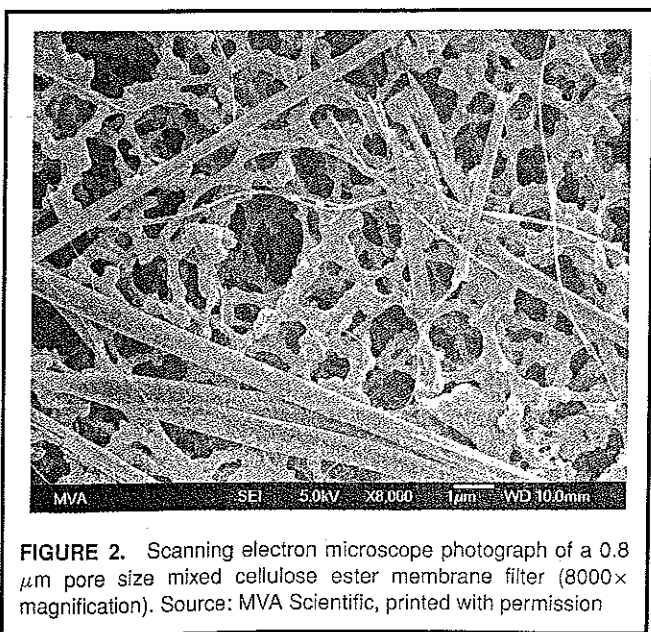


FIGURE 2. Scanning electron microscope photograph of a 0.8 μm pore size mixed cellulose ester membrane filter (8000 \times magnification). Source: MVA Scientific, printed with permission

filter assembly for 0.45 μm pore size MCE as well as for the 0.4 μm pore size PC filters.⁽³⁾ It is important to note that the NIOSH phase contrast method (PCM) imposes no such requirement, nor does it require low temperature ashing of the filter. Therefore, the results of the present study apply to TEM but not directly to PCM analysis.

Studies reporting the fiber retention efficiencies of MCE and PC membrane filters for asbestos aerosols are meager. One study investigated the fiber retention efficiencies of 8 μm pore size MCE filters and 0.2, 0.4, and 0.8 μm pore size PC filters for aerosols of chrysotile asbestos.⁽¹²⁾ For MCE filters with 8- μm pores, the fiber retention efficiency at a face velocity of 3.5 cm/s fell from 100% for fibers longer than 5 μm to 75% for fibers longer than or equal to 2 μm , and to 25% for fibers approximately 0.5 μm long. For PC filters with pore diameters of 0.2, 0.4, and 0.8 μm , fiber retention efficiencies began to drop for fiber lengths shorter than 3 μm and fiber diameters smaller than 0.2 μm . For PC filters with 0.2- μm pores, the fiber retention efficiencies for fibers longer than 0.5 μm did not drop below approximately 80%, whereas for 0.8 μm pores, the fiber retention efficiencies dropped to near zero for fiber lengths below 0.5 μm and diameters below 0.05 μm .

This study showed that fiber retention efficiencies decrease substantially with fiber length for both MCE and PC pore filters of larger pore size. In addition, the orientation of the airborne fibers as they approach the filter pore entrances may have an important effect on their ability to penetrate the filter.

A literature review⁽¹³⁾ did not identify any study that compared the fiber retention efficiencies of 0.45 μm and 0.8 μm pore size MCE or 0.4 μm pore size PC membrane filters for asbestos aerosols. Information culled from an informal survey⁽¹³⁾ of asbestos analytical laboratories, members of the American Society for Testing and Materials (ASTM) and Environmental Information Association (EIA) revealed that MCE filters were used primarily for airborne asbestos sampling. Accordingly, it was concluded that testing of the PC filters would not be conducted in this study, allowing the project to concentrate its efforts and funding on 0.45 μm and 0.8 μm pore size MCE filters that are widely used in asbestos exposure studies today.

Therefore, this study generated chrysotile asbestos fibers (both shorter and longer than 5 μm) in an aerosol chamber and collected the fibers by 25 mm diameter MCE filter media to compare the efficiency of 0.45 μm pore size vs. 0.8 μm pore size filter media. In addition, the effect of plasma etching times on fiber densities was evaluated.

The goal of this research study was to determine the effect of mixed cellulose ester membrane filter pore size on retention of asbestos fiber aerosols in the filter matrix and the ability to identify and count these fibers by TEM analysis. The following are the specific objectives of this study:

- Compare the fiber retention efficiencies (structures longer than or equal to 0.5 μm) of asbestos aerosols of 0.45 μm and 0.8 μm pore size mixed cellulose ester filters.

- Compare fiber retention efficiencies (structures longer than 5 μm) of asbestos aerosols of 0.45 μm and 0.8 μm pore size mixed cellulose ester filters.
- To evaluate the effect of plasma etching time (2, 4, 6, 8, and 16 min) on 0.45 μm pore size mixed cellulose ester filters on total asbestos density (structures longer than or equal to 0.5 μm).
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METHODS

Preparation of Samples for Analysis

SRI International (SRI) prepared 25 mm diameter (0.45 μm and 0.8 μm pore size) MCE filters with chrysotile asbestos in an aerosol chamber. Chrysotile was chosen for the study because it provides a more rigorous test of the analytical method than do the amphiboles. Chrysotile fibers are usually thinner than amphibole fibers and are more difficult for the analyst to detect. The filters were prepared at two fiber loading levels: "low" nominal loading (2–5 fibers per grid opening, which equates to 200–500 s/mm²) and "high" nominal loading (greater than 5 fibers per grid opening, which equates to greater than 500 s/mm²). The filters were prepared in four batches of 18 filters each as shown in Table I.

Dust Generation and Collection System

Test atmospheres of dusts and fibers were dynamically generated in SRI's dust generation and collection system. The airstreams pass through ionizers to prevent static charge effects. The system consists of the following components:

- Dust feeder, which delivers a continuous stream of aerosol material.
- Sonic velocity disperser, which disperses, de-agglomerates, and dilutes the aerosol.
- Settling tower, where large particles are removed.
- Sample collection chambers, where 320 samples can be collected simultaneously.

TABLE I. Batch Setup for 25-mm MCE Filter Loading Experiment

Batch	Filter Pore Size/Loading and Number of Samples			
	0.45 μm		0.8 μm	
	Low	High	Low	High
1	12	—	6	—
2	—	6	—	12
3	6	—	12	—
4	—	12	—	6
Total	18	18	18	18

Dust Feeder

The asbestos-containing powder was metered into the collection system by a grooved disk, which rotates at a known rate. The powder is pneumatically unloaded from a groove in the disk and then conveyed to a sonic velocity disperser.

Powder is loaded into the top of the powder hopper through the powder feed port. The powder then drops down into the hopper connector where it is pushed into the groove of the disk by rubber wipers attached to the bottom of the agitator shaft. A spring-loaded guard ring surrounds the hopper connector and scrapes the disk to prevent it from carrying away excess powder. The rotation of the disk continuously carries the powder in the groove to the unloading nozzle where it is removed pneumatically by compressed air. The powder feed rate is determined entirely by the rotation speed of the disk and the size of the groove. The loading of powder on sample filters is further adjusted by varying the collection time.

Asbestos fiber atmospheres are generated using a two-component fluidized bed consisting of bronze powder and sized asbestos fibers. The bronze particle nominal size is 0.2 μm . The supporting data for the homogenous atmosphere are the density of asbestos on the receiving MCE filters. Fibers were sized using ISO 10312 method on a calibrated etched TEM screen. The flow rate is 2 L/min. Because the study objectives were to determine fiber retention efficiencies of fibers, the airflow duration is not relevant. Rather, the crucial measurement endpoints are the resultant fiber density on the filters and the measured variability among filters.

By proper adjustment of airflow through the bottom and across the top of the bed, the bronze powder bed is fluidized and the asbestos fibers are stripped at a low rate and fed to the sonic velocity disperser. There is a concentration gradient using this system because the asbestos is depleted from the fluidized bed. However, because a homogeneous atmosphere is produced, each sampling port still collects an equivalent amount of asbestos. By varying the sampling time, the asbestos loading on the cassettes is adjusted. By using a combination of the fluidized bed and the powder feeder, a variety of fibers and particles is loaded onto a filter.

Sonic Velocity Disperser and Settling Tower

The airstream from the dust feeder carries the aerosol to the sonic velocity disperser. Dilution air is also delivered to the sonic velocity disperser where it de-agglomerates the aerosol under the action of an online static eliminator and high air velocity. The aerosol then enters the settling tower, the linear velocity is reduced, and the larger particles settle out to the base of the settling tower. The diluted aerosol is then divided uniformly among the four collection chambers.

Sample Collection Chamber

The base section of the sample collection chamber consists of layers of gaskets and machined-aluminum sheets. Eighty sampling ports are situated in an 8 \times 10 matrix arrangement. Downstream from each port is a critical flow orifice. The mounting sheet in which the 80 critical flow orifices are

embedded forms the upper section of a vacuum chamber, so that a vacuum to this chamber creates the necessary pressure differential to operate the orifices. Aerosol enters the collection chambers through 20 symmetrically located passages. The 320 orifices (80 for each of four sample collection chambers) all have the same diameter and were calibrated at the time the system was constructed to ensure that all the ports sample at the same flow rate. The orifices form a matched set, with a maximum flow rate of 2 L/min through each air monitor in the system. SRI collected 100 filters in each batch and used 80 of the primary filter cassettes (e.g., in a 0.45 μm pore size batch the 0.45 μm pore size filters are the primary filter cassettes) and 20 of the secondary filter cassettes (e.g., in a 0.45 μm pore size batch the 0.8 μm pore size filters are the secondary filter cassettes), so that the variable of different batches could be adequately measured and controlled. The 80 primary filter cassettes and 20 secondary filter cassettes were divided evenly between the four quadrants of the chamber.

The collection chambers are opened from the top by removing a cover. Air monitoring filter cassettes are connected to the sampling port using a luer fitting. Quality control activities include checking each orifice flow rate with a digital flow meter before and after sample generation and analyzing for background levels to prevent carryover contamination.

Sample Analysis Strategy

Fiber Retention Efficiency of 0.45 μm and 0.8 μm Pore Size MCE Filters

Seventy-two filter samples were prepared and analyzed to test pore size differences and fiber loading differences between the two MCE filter types (Table I). Eighteen filters were analyzed for each of four batches. Twelve of the primary filters and six of the secondary filters were analyzed for each batch.

Effect of Plasma Etching Time on Asbestos Density

Annex A "Determination of Operating Conditions for Plasma Asher" of ISO Method 10312:1995 requires etching of collapsed filters for 8 min using operating parameters determined for completely ashing uncollapsed filters in 15 min. Including the specified 8-min etching time, three additional etching times were used to etch the 0.45 μm MCE filters. Hence, a total of 12 filters were etched for each of four different times (2, 4, 8, and 16 min). The filters were loaded at a "high" nominal loading.

Analytical Methodology

TEM Specimen Preparation

TEM specimens were prepared from the air filters using the dimethylformamide (DMF) collapsing procedure of ISO 10312:1995, as specified for cellulose ester filters. DMF was used as the solvent for dissolution of the filter in the Jaffe washer. Prior to etching the filters, a March Plasmod asher was calibrated in accordance with ISO 10312:1995 procedures whereby an uncollapsed filter was oxidized under controlled settings in approximately 15 min. After asher calibration,

the filters were prepared using ISO 10312:1995 procedures and etched for either 2, 4, 8, or 16 min. For each filter, an equal number of grid openings were examined on at least two prepared TEM specimen prepared from a one-quarter sector of the filter using 200 mesh-indexed copper grids. The remaining part of the filter was archived in the original cassette in clean and secure storage.

The minimum aspect ratio for the analyses was 3:1, as permitted by ISO 10312:1995. As required in the ISO method, any identified compact clusters and compact matrices were counted as total asbestos fibers, even if the 3:1 aspect ratio was not met. All fibers longer than or equal to 0.5 μm were quantified with the following breakdown according to ranges by length: (a) longer than or equal to 0.5 to 5.0 μm ; (b) longer than 5.0 to 10.0 μm ; and (c) longer than 10.0 μm . The fiber counting data were distributed approximately equally among a minimum of two specimen grids prepared from different parts of the filter sector. The TEM specimen examinations were performed at approximately 20,000 \times magnification. Phase contrast microscopy equivalent asbestos structures (PCME) were also determined. PCME asbestos structures, as defined by ISO 10312:1995, are longer than 5 μm and from 0.2 to 3.0 μm in diameter with an aspect ratio of 3:1 or greater.

The analytical sensitivity was greater than or equal to 9.1 asbestos structures per square millimeter (s/mm^2), calculated as 1 structure/10 grid openings of $0.011 \text{ mm}^2 = 0.091$. In principle, any analytical sensitivity can be achieved by increasing the number of grid openings or fields examined. Likewise, statistical uncertainty around the number of fibers observed can be reduced by counting more fibers. Stopping rules are needed to identify when microscopic examination should stop, both at the low end (zero or very few fibers observed) and at the high end (many fibers observed). The analysis was terminated upon completion of counting greater than or equal to 25 asbestos structures in a minimum of 10 grid openings or 100 asbestos structures in 4 grid openings. In any case, completion of the grid opening being analyzed when the stopping rules have been met was completed.

The filter samples generated by SRI were monitored for absolute loading and for intra-batch uniformity by an independent quality control (QC) laboratory, RTI International, which prepared and analyzed samples and provided feedback to SRI regarding filter loading so that the batches met the target densities. They also used the data to validate the uniformity of density of filters within each batch. For each batch of filters produced, a relative standard deviation (RSD) of fibers per grid opening was developed with 40 grid openings analyzed. Based on historical RSD levels for SRI filters, each batch was expected to have an RSD at or below 0.50, which was the case for this study.

Before filter samples were loaded with chrysotile asbestos, two unused filters from each filter lot of 0.45 and 0.8 μm , filters were analyzed by the QC laboratory to determine the mean asbestos structure count. The lot blanks were analyzed for asbestos structures by using ISO 10312:1995. In all cases, the mean count for all types of asbestos structures was less than

TABLE II. Interlaboratory Duplicates Analysis of MCE Filters for Chrysotile Asbestos by TEM

Sample No.	Laboratory	Analyses		Actual Variability	Accepted Variability
		# Structures	Structures/mm ²		
A0611022-001A	Primary	26	230	1.8	2.24
	QC	23	270		
A0611022-002A	Primary	28	250	1.3	2.24
	QC	19	220		
A0611022-003A	Primary	34	300	0.39	2.24
	QC	27	310		
A0611022-004A	Primary	31	280	0.86	2.24
	QC	22	280		

$$\text{Analytical Variability} = \frac{|(\text{Analysis A}) - (\text{Analysis B})|}{\sqrt{\text{Analysis A} + \text{Analysis B}}}$$

This variability is an estimate of the standard deviation of the difference based on a Poisson counting model. The value 2.24 was selected as targeting false positive rates of 2.5% (1/40) for the Poisson model.

or equal to 18 s/mm². Laboratory blanks are unused filters that are prepared and analyzed in the same manner as the field samples to verify that reagents and equipment are free of the subject analyte and that contamination has not occurred during the analysis process. The laboratory analyzed two 0.45 μm and two 0.8 μm pore size MCE filters. Blanks were prepared and analyzed along with the other samples. Asbestos was not present on any of the samples at an analytical sensitivity of 9.1 s/mm².

After analysis by the primary laboratory, selected filters and grid preparations were sent to the QC laboratory for analysis as an independent QA/QC check. The QA/QC sample analyses included duplicates and verified counts by TEM. The duplicate analyses were conducted by re-preparing and analyzing the same filter using the same ISO 10312:1995 counting rules. Results of the QC duplicate analysis are presented in Table II. In Table III, the third column lists the number of structures analyzed, and the fourth column lists the density of asbestos structures per unit area. Note that the primary laboratory used a grid opening size of 0.011 mm², and the QC laboratory used a grid opening size of 0.0086 mm². Column 5 presents the results of the duplicate sample variability. All four interlaboratory duplicate samples met the acceptance criteria.

Verified Fiber Counts

Verification counting involved re-examination of the same grid openings analyzed by the primary laboratory. The verification counting was performed on two of the analyses for each of the filter pore sizes. Verified counting was conducted using the procedure defined in NISTIR 5351, "Airborne Asbestos Method: Standard Test Method for Verified Analysis of Asbestos by Transmission Electron Microscopy – Version 2.0."

Results of interlaboratory QC verified counting by TEM are presented in Table III. The third column gives the total number of asbestos structures counted in the specified grid openings that were determined to be true positives (TP). Column 4 gives the number of false positives (FP), and Column 5 gives the number of false negatives (FN). The results of all four analyses are combined at the bottom, and ratios of true positives, false positives, and false negatives are developed in the final two rows for both the primary and QC laboratory. Column 6 shows the "pass" (Yes) or "fail" (No) status of the comparison. The acceptable variability is greater than 80% true positives, less than 20% false negatives, and less than 20% false positives. All interlaboratory verified count analysis met the acceptance criteria.

TABLE III. Interlaboratory Verified Count Analysis of MCE Filters for Chrysotile Asbestos by TEM

Sample No.	Laboratory	Number of Structures			Pass?
		True Positive	False Positive	False Negative	
A0611024-003A	Primary	24	0	1	
	QC	25	1	0	
A0611024-002A	Primary	35	3	0	
	QC	32	0	3	
A0611024-004A	Primary	5	0	1	
	QC	6	1	0	
A0611024-005A	Primary	8	0	0	
	QC	7	0	1	
Totals	Primary	72	3	2	
	QC	70	2	4	
Percentages	Primary	97%	4%	3%	Yes
	QC	95%	3%	5%	Yes

TABLE IV. Mean Chrysotile Asbestos Density (s/mm²) by Batch and Fiber Length

Batch	Mean Fiber Density (s/mm ²) by Length of Fibers (standard deviation)					
	Filter Low Loading			Filter High Loading		
	≥ 0.5 – 5 μm	>5–10 μm	>10 μm	≥ 0.5 – 5 μm	>5–10 μm	>10 μm
0.45 μm pore size						
1	237 (113)	63 (22)	21 (112)	—	—	—
2	—	—	—	585 (92)	284 (86)	89 (30)
3	317 (41)	71 (21)	21 (14)	—	—	—
4	—	—	—	1200 (296)	235 (91)	66 (48)
0.8 μm pore size						
1	194 (44)	60 (21)	19 (13)	—	—	—
2	—	—	—	429 (80)	225 (76)	88 (33)
3	288 (69)	78 (30)	22 (10)	—	—	—
4	—	—	—	960 (299)	261 (70)	71 (27)

RESULTS AND DISCUSSION

Fiber Retention Efficiency

A total of 72 filters (18 of 0.45 μm pore size and 18 of 0.8 μm pore size) were loaded with asbestos at two filter loadings (low = 2–5 fibers/grid opening; and high = greater than 5 fibers/grid opening). The experiment was conducted in four batches of 18 filters each (Table I).

The number of filters provided a sufficient number of samples consistent with the time, funding, and availability of the aerosol chamber, while still meeting the study objectives. Ideally, numerous asbestos and asbestiform fibers would be analyzed. However, to ensure intra-batch and intra-sample comparability, chrysotile fibers were generated exclusively. Because it is the most common fiber type in most asbestos exposure scenarios to date, and owing to its finely fibrous nature, it is also the ideal form of asbestos to study post-preparation fiber retention in filters. Lee and Liu's⁽¹⁴⁾ equation for predicting most penetrating particle diameter ($d_{p,min}$) is:

$$d_{p,min} = 0.885 \left[\left(\frac{K}{1-\alpha} \right) \left(\frac{\sqrt{\lambda kT}}{\eta} \right) \left(\frac{d_f^2}{U} \right) \right]^{2/9}$$

where K is the hydrodynamic factor, α = solidity of filter (1 – porosity), λ is the mean free path of the gas molecules, k is the Boltzmann constant, T is the absolute temperature, η is the air viscosity, d_f is the filter fiber diameter, and U is average air velocity inside the filter medium. Therefore, the most penetrating particle diameter decreases with decreasing pore size in the filter medium. This relationship holds for both fibrous and membrane filters,⁽¹⁵⁾ and in porous-membrane filters.⁽¹⁶⁾

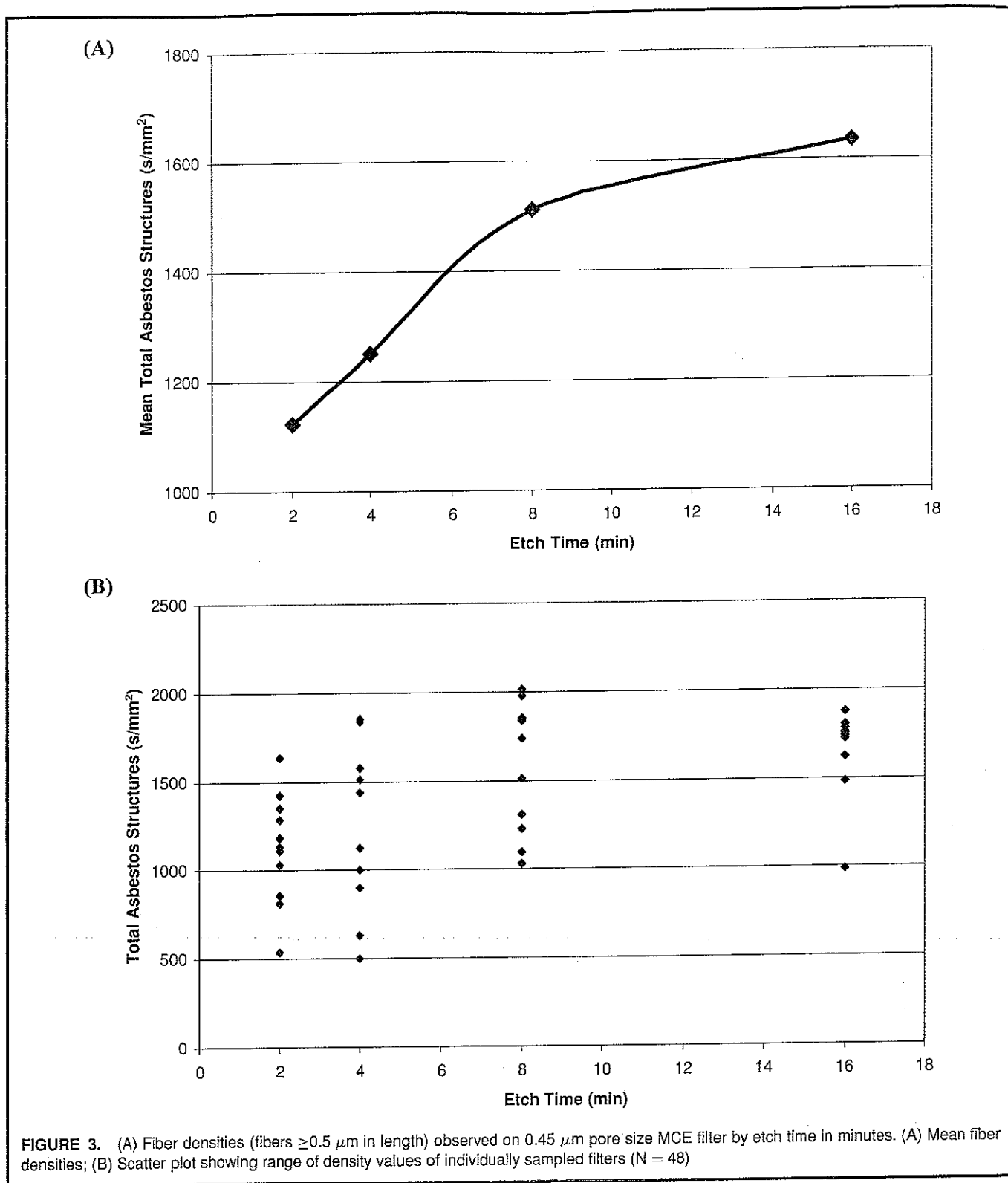
Of all asbestos fiber types, chrysotile is the most likely to penetrate the tortuous matrix of MCE filter material, thus optimizing the ability of the study to employ electron microscopy to characterize differences in asbestos post-preparation fiber

retention due to MCE pore size (larger pore sizes equate to greater potential penetration of fibers into the matrix) and due to differential plasma etching time. Amphibole asbestos fibers, with their larger average diameter and length⁽¹⁷⁾ are less likely to penetrate the MCE matrix and, therefore, are more easily visible than most chrysotile fibers by electron microscopy. In addition, since chrysotile asbestos is by far the most commonly seen asbestos type on air filters (such as from abatement sites), it best reflects real-world situations. Thus, these results for chrysotile asbestos provide an indication of TEM analytical effectiveness for numerous fibers, including amphibole asbestos.

All asbestos structures greater than or equal to 0.5 μm were quantified and categorized according to three ranges by length: longer than or equal to 0.5 to 5 μm; longer than 5 to 10 μm; and longer than 10 μm. The asbestos fiber distribution for the low and high filter loadings is presented in Table IV. It is important to note that the sample sizes for individual batch comparisons are too small to give statistically significant results (except for Batch 2), even though the fiber retention efficiency of the 0.45 μm filter was greater in each case. Using the sum of the Wilcoxon statistics increases the effective sample size for the comparison between pore sizes. The Wilcoxon test is a nonparametric procedure that does not depend on comparisons of means between batches. Rather, it uses the ranking of the observations in the combined data from the two batches and as such is not particularly sensitive to variability that might affect a comparison of means.

Any variation in airborne asbestos concentrations, in time or space, is reflected in the variability of the individual fiber counts and is accounted for by the pairing of the data within batches in the statistical analysis. The nonparametric approach also mitigates any variability in the environment.

The mean filter density (total asbestos structures per mm²) for the two filter types in each batch is presented in Tables V and VI. In each batch, the mean density on the 0.45 μm filters is higher than on the 0.8 μm filters. In Batch 2, the



difference is statistically significant using both the two-sample t-test ($p = 0.008$) and the nonparametric Wilcoxon Rank-Sum test ($p = 0.01$). In the other three batches, the difference is not statistically significant.

The two "low loading" batches differ substantially, as do the two "high loading" batches. As evidence, the $0.8 \mu\text{m}$ density in Batch 3 is higher than the $0.45 \mu\text{m}$ density in Batch 1, even though both batches were loaded at the same nominal

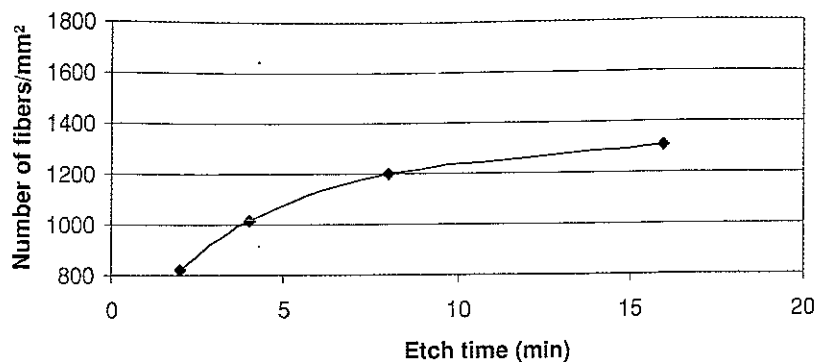


FIGURE 4. Mean fiber densities (fibers ≥ 0.5 to $5 \mu\text{m}$ in length) observed on $0.45 \mu\text{m}$ pore size MCE filter at varying etching times

level. Likewise, the $0.8 \mu\text{m}$ density in Batch 4 is higher than the $0.45 \mu\text{m}$ density in Batch 2. In fact, the between-batch differences (at the same nominal loading) are greater than the differences between the two filter types. Thus, it is not appropriate to combine the four batches into a single dataset for purposes of an overall comparison between the two filter types.

To make the overall comparison, the sum of the Wilcoxon statistics for the four separate batches was used. In each batch, the Wilcoxon statistic is the rank-sum for the $0.45 \mu\text{m}$ densities in the 18 samples comprising the batch. Under the null hypothesis that the two filter types have the same fiber retention efficiency, this statistic has (approximately) a normal distribution with mean 57 (Batches 2 and 3) or 114 (Batches 1 and 4), and variance 114 (all batches). Thus, under the null hypothesis, the sum of the four Wilcoxon statistics is approximately normal with mean 342 and variance 456. The observed value of the sum is 395.5, resulting in a test statistic $z = (395.5 - 342) / 21.4 = 2.50$, with a p-value of 0.01. Thus, the null hypothesis is rejected, and it is concluded that the fiber retention efficiency of the $0.45 \mu\text{m}$ pore size filter for fibers longer than or equal to $0.5 \mu\text{m}$ is significantly higher than that of the $0.8 \mu\text{m}$ pore size filter. However, for fibers longer than $5 \mu\text{m}$ there is no difference in the two filter pore sizes.

Effect of Plasma Etching Time on Total Asbestos Density

Four different etching times were used to etch $0.45 \mu\text{m}$ filters. A total of 12 filters were etched for each of the four times (2, 4, 8, and 16 min). The filters were loaded in at the "high" nominal loading. Table VI shows the mean total asbestos (chrysotile) loading (s/mm^2) for fibers longer than or equal to $0.5 \mu\text{m}$ for each etching time.

The mean density increases with etching time. To examine the relationship between etching time and density, two regression models were fit to the data. The first model assumes a linear relationship between etching time (t) and density (TA):

$$\text{TA} = a + b * t$$

The fitted equation was

$$\text{TA} (\text{s}/\text{mm}^2) = 1113 + 35.7 * t$$

$$(R^2 = 0.24)$$

The relatively low value of R^2 is due to the considerable variability in densities observed at each etching time. However, the coefficient of t is highly significant ($\text{SE} = 9.27$). This

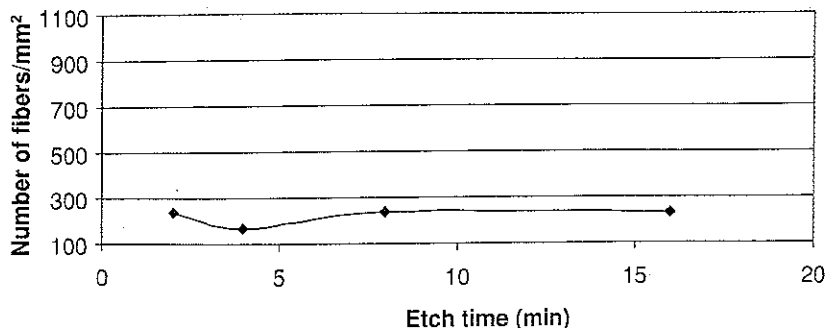


FIGURE 5. Mean fiber densities (fibers 5 to $10 \mu\text{m}$ in length) observed on $0.45 \mu\text{m}$ pore size MCE filter at varying etching times

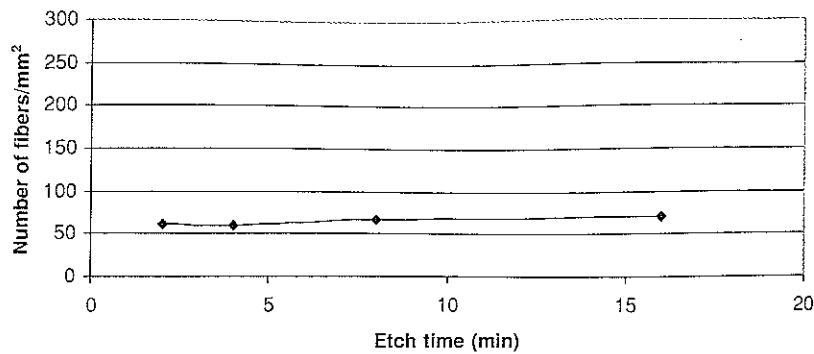


FIGURE 6. Mean fiber densities (fibers 5 to 10 μm in length) observed on 0.45 μm pore size MCE filter at varying etching times

regression indicates that each additional minute of etching time adds an average of 35.7 s/mm^2 to the total asbestos density.

The second regression model assumes a logarithmic relationship between density and etching time, of the form:

$$\text{TA} = a + b * \ln(t)$$

The fitted equation is:

$$\text{TA} (\text{s}/\text{cm}^2) = 931 + 259 * \ln(t)$$

$$(R^2 = 0.27)$$

R^2 is slightly higher than for the linear model. Again, the regression is highly significant (SE of coefficient = 63). Doubling the etching time increased the asbestos filter loading in this study by an average of 13%.

Based on physical limitations, greater number of fibers observed with increased etching time will eventually diminish, i.e., there is a level of etching time beyond which no increase in density is expected (Figures 3 and 4). The data from this experiment do not appear to shed light on what this level might be. For example, the increase in density from 8 to 16 min is comparable to that from 2 to 4 min. However, the increase in density with etching time does not appear to be the case

for fibers longer than 5 μm (Figures 5 and 6). These data suggest that the etching time of 8 min that is specified in ISO 10312:1995 is adequate for fibers longer than 5 μm . If fibers less than 5 μm are of interest, additional research may be needed to determine the optimum etching time.

The mean filter density (total asbestos structures, longer than 5 μm , per mm^2) for the two filter types in each batch is shown in Table VII.

In Batches 1 (low loading) and 2 (high loading), the mean density on the 0.45 μm filters is higher than on the 0.8 μm filters. In Batches 3 (low loading) and 4 (high loading), the reverse is true; i.e., the 0.8 μm filters are higher. None of the differences are statistically significant using both the two-sample t-test. When Batches 1 and 3 (low loading) and Batches 2 and 4 (high loading) are combined, the differences between the filter types are even smaller. It is concluded that for fibers longer than 5 μm , there is no difference between the fiber retention efficiencies of the 0.45 μm and 0.8 μm filters.

Table VIII shows that for fibers longer than 5 μm (s/mm^2) the mean densities for the 2-, 8-, and 16-min etching times are virtually identical. The mean density for the 4-min etching time is slightly lower.

To examine the relationship between etching time and density, two regression models were fit to the data. The first model assumes a linear relationship between etching time (t) and density (TA):

$$\text{TA} = a + b * t$$

TABLE V. Mean Total Chrysotile Asbestos Density (s/mm^2) by Batch and Filter Type

Batch	Mean Fiber Density (s/mm^2) by Filter Pore Size and Nominal Loading (standard deviation)			
	0.45 μm		0.8 μm	
	Low Loading	High Loading	Low Loading	High Loading
1	321 (114)	—	274 (57)	—
2	—	958 (170)	—	743 (125)
3	413 (55)	—	388 (89)	—
4	—	1512 (369)	—	1304 (336)

TABLE VI. Mean Total Chrysotile Asbestos (fibers $>0.5 \mu\text{m}$) Density (s/mm^2) for Variable Etching Times (standard deviation)

Filter Loading	Plasma Etching Time for 0.45 μm MCE Filters (min)			
	2	4	8	16
High	1123 (295)	1251 (442)	1512 (369)	1635 (236)

TABLE VII. Mean Chrysotile Asbestos Density (s/mm²) for Fibers ≥ 5 μm in Length by Batch and Filter Type (standard deviation)

Batch	Mean Fiber Density (s/mm ²) by Filter Pore Size and Nominal Loading			
	0.45 μm		0.8 μm	
	Low Loading	High Loading	Low Loading	High Loading
1	84 (30)	—	80 (28)	—
2	—	373 (102)	—	313 (104)
3	92 (33)	—	100 (32)	—
4	—	301 (118)	—	333 (85)

The fitted equation is:

$$TA \text{ (s/mm}^2\text{)} = 267 + 2.2 * t$$

$$(R^2 = 0.016)$$

The regression is not statistically significant.

The second regression model assumes a logarithmic relationship between density and etching time of the form:

$$TA = a + b * \ln(t)$$

The fitted equation is:

$$TA \text{ (s/mm}^2\text{)} = 266 + 10.5 * \ln(t)$$

$$(R^2 = 0.007)$$

Again, the regression is not statistically significant. The fact that neither regression is statistically significant indicates that, for 0.45 μm filters, there is no statistically significant relationship between etching time and density of fibers longer than 5 μm.

This is consistent with a study conducted by Chatfield.⁽¹¹⁾ The study showed that fiber densities for fibers longer 5 μm are similar for 0.2 μm pore size PC filters and various etching schedules for 0.22 μm pore size MCE filters. In particular, plasma etching had no effect on the reported fiber densities of fibers longer than 5 μm. At the 1% significance level, there

TABLE VIII. Mean Chrysotile Asbestos Density for Fibers > 5 μm (s/mm²) for Variable Etching Times (standard deviation)

Filter Loading	Plasma Etching Time for 0.45 μm MCE Filters (min)			
	2	4	8	16
High	301 (75)	232 (99)	301 (118)	303 (77)

were no statistically significant differences between the mean fiber densities for any of the etching preparations evaluated.

CONCLUSIONS

The null hypothesis is that the two mixed cellulose ester (MCE) filter types (0.45 μm and 0.8 μm pore size) have the same fiber retention efficiency for asbestos aerosol (structures longer than or equal to 0.5 μm length). The null hypothesis was rejected, and it is concluded the fiber retention efficiency of the 0.45 μm pore size MCE filter is statistically significantly higher than that of the 0.8 μm pore size MCE filter (p = 0.01) for fibers longer than or equal to 0.5 μm. However, for asbestos structures longer than 5 μm, there is no statistically significant difference between the fiber retention efficiencies of 0.45 μm and 0.8 μm pore size MCE filters (p > 0.05).

There is a significant difference in the effect of etching times for fibers shorter than 5.0 μm and fibers longer than 5.0 μm. The mean density of asbestos fibers longer than 0.5 μm increases with etching time (2, 4, 8, and 16 min) of 0.45 μm pore size MCE filters. Regression analysis of etching time and densities showed that doubling the etching time adds an average of the 13% to the total asbestos density. This increase is a diminishing percentage of the total fiber count as the etching time increases, e.g., 20% at 2 min, and 12% at 8 min. There is likely an etching time beyond which no increase in density is expected and, in fact, would decrease; the data from this experiment did not identify this etching time. However, etching the filter for longer periods may remove too much filter so that a specimen for TEM analysis cannot be prepared. For fibers longer than 5.0 μm, there is no significant difference in numbers of structures counted at the etching times used in these tests.

RECOMMENDATIONS

This research study demonstrates that the fiber retention efficiency of a 0.45 μm pore size MCE filter for aerosols of asbestos fibers (structures longer than or equal to 0.5 μm) is greater than that for a 0.8 μm pore size MCE filter. However, there is no difference in fiber retention efficiency between these pore sizes for structures longer than 5.0 μm. If the exposure study is focused on fibers less than 5.0 μm, the investigator should use filters with 0.45 μm pore size. If the exposure study is interested only in structures longer than 5.0 μm, then either filter pore size may be used.

Because most asbestos exposure risk models include fibers longer than 5.0 μm, the 8-min etching time specified in ISO 10312:1995 is adequate. However, if an exposure study is focused on fibers less than 5.0 μm, the etching time of 8 min should be reviewed. A study should be conducted to determine the etching time beyond which no significant increase in asbestos density of fibers less than 5.0 μm is expected.

NIOSH Method 7402 notes that a 0.45 μm pore size filter may be difficult to use with some personal sampling pumps due to the pressure drop across this filter. The tandem MCE filter

assembly (0.45 μm pore size collection filter and 5 μm pore size diffusing filter) recommended by AHERA (40 CFR §761), ISO Method 10312:1995, and ASTM Method D 6281-04 may preclude the use of some battery-powered personal sampling pumps due to the resultant high pressure drop. Analysis of filters by TEM requires the use of the 5 μm pore size diffusing filter to ensure uniform deposition on the primary collection filter. A study should be conducted to evaluate the difference between asbestos aerosols collected on 0.45 μm and 0.8 μm pore size MCE filters with and without the 5 μm pore size MCE diffusing filter. Also, specifications for personal pumps should be investigated to determine optimum requirements for sampling using the 0.45 μm pore size collection filter and 5 μm pore size diffusing filter combination.

This study has focused on MCE filters because this filter type is the primary choice for air monitoring. Exposure to asbestos through inhalation is considered the most likely route for asbestos exposure. Polycarbonate (PC) filters are used in monitoring asbestos in water and possibly by some studies of inhalation. Because no data have been found comparing the relative effectiveness of MCE and PC filters, research should be considered to compare the retention of asbestos fibers on 0.45 μm pore size MCE filters to 0.4 μm pore size polycarbonate filters.

Chrysotile asbestos was chosen for this study because it provides a more rigorous test of the analytical method than the more readily observed amphiboles. An additional comparison of filter efficiency should be made using amphibole materials to ensure the findings of this study apply to amphibole fibers as well. Also, because only asbestos fibers were used in this study it would be appropriate to include a suitable particle matrix in future testing.

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REFERENCES

1. "Appendix A to Subpart E of Part 763. Interim Transmission Electron Microscopy Analytical Methods—Mandatory and Nonmandatory—and the Mandatory Section to Determine Completion of Response Action," *Code of Federal Regulations Title 40*, Part 763, 1987.
2. **ASTM International**: D 6821 Airborne asbestos concentration in ambient and indoor atmospheres as determined by transmission electron microscopy direct transfer (TEM). In *ASTM Standards on Indoor Air Quality*, Second Edition. West Conshohocken, Pa.: ASTM International, 2002.
3. **International Organization for Standardization (ISO)**: *Ambient Air—Determination of Asbestos Fibres—Direct-Transfer Transmission Electron Microscopy Method (ISO 10312)* [Standard]. Geneva: ISO, 1995.
4. **International Organization for Standardization (ISO)**: *Ambient Air—Determination of Asbestos Fibres—Indirect-Transfer Transmission Electron Microscopy Method (ISO 13794)* [Standard]. Geneva: 1999.
5. **National Institute for Occupational Safety and Health (NIOSH)**: Asbestos by TEM, method 7402. In *NIOSH Manual of Analytical Methods (NMAM)*, 4th ed., P.C. Schlecht and P.F. O'Connor (eds.). Cincinnati, Ohio: NIOSH, 1994.
6. **U.S. Environmental Protection Agency (USEPA)**: *Environmental Asbestos Assessment Manual Superfund Method for the Determination of Asbestos in Ambient Air, Part 1: Method (EPA 540/2-90-005a)*. Washington, D.C.: USEPA, May 1990.
7. **U.S. Environmental Protection Agency (USEPA)**: *Methodology for the Measurement of Airborne Asbestos by Electron Microscopy* by G. Yamate, S.C. Agarwal, and R.D. Gibbons. Draft Report, EPA Contract 68-02-3266 for Environmental Monitoring Systems Laboratory, Office of Research and Development, Research Triangle Park, N.C.. Washington D.C., USEPA, 2002.
8. **National Institute for Occupational Safety and Health (NIOSH)**: Asbestos and other fibers by PCM, method 7400. In *NIOSH Manual of Analytical Methods (NMAM)*, 4th ed., P.C. Schlecht and P.F. O'Connor (eds.). Cincinnati, Ohio: NIOSH, 1994.
9. "Asbestos in Air, Method Number ID-160." [Online] Available at <http://www.osha.gov/dts/sltc/methods/toc.html> Accessed June 1, 2008.
10. **International Organization for Standardization (ISO)**: *Air Quality—Determination of the Number Concentration of Airborne Inorganic Fibers by Phase Contrast Microscopy—Membrane Filter Method (ISO 8672)* [Standard]. Geneva: ISO, 1993.
11. **Chatfield, E.J.**: Measurements of chrysotile fiber retention efficiencies for polycarbonate and mixed cellulose ester filters. In *Advances in Environmental Measurement Methods for Asbestos, American Society for Testing and Materials (STP 1342)*, M.E. Beard and Harry L. Rook (eds.). West Conshohocken, Pa.: ASTM, 2000.
12. **Spurny, K.**: On the filtration of fibrous aerosols. *J. Aerosol. Sci.* 16(3):450-455 (1986).
13. **U.S. Environmental Protection Agency (USEPA)**: *Sampling and Analysis of Asbestos Fibers on Filter Media to Support Exposure Assessments: Scoping Effort* by M.E. Beard and J.R. Kominsky. Report prepared by Environmental Quality Management, Inc., Cincinnati, Ohio, and RTI International, Research Triangle Park, N.C. for U.S. EPA National Exposure Research Laboratory (Task Order No. 0020, Contract No. 68-C-00-186). Washington, D.C.: USEPA, 2006.
14. **Lee, K.W., and B.Y.H. Liu**: On the minimum efficiency and the most penetrating particle size for fibrous filters. *J. Air Pollut. Control Assoc.* 30:377-381 (1980).
15. **Rubow, K.L.** "Submicrometer Aerosol Filtration Characteristics of Membrane Filters." Ph.D. diss., University of Minnesota, Minneapolis, Minn. (1981).
16. **Baron, P.A., and K. Willeke**: *Aerosol Measurement: Principles, Techniques, and Applications*, Second Edition. Hoboken, N.J.: Wiley-Interscience, 2005. pp. 213-215
17. **Wylie, A.G., R.L. Virta, and E. Russek**: Characterizing and discriminating airborne amphibole cleavage fragments and amosite fibers: Implications for the NIOSH method: *Am. Ind. Hyg. Assoc. J.* 46:197-201 (1985).